

# Can agriculture be eco-friendly?

## Plant extracts as grapevine defense inducers

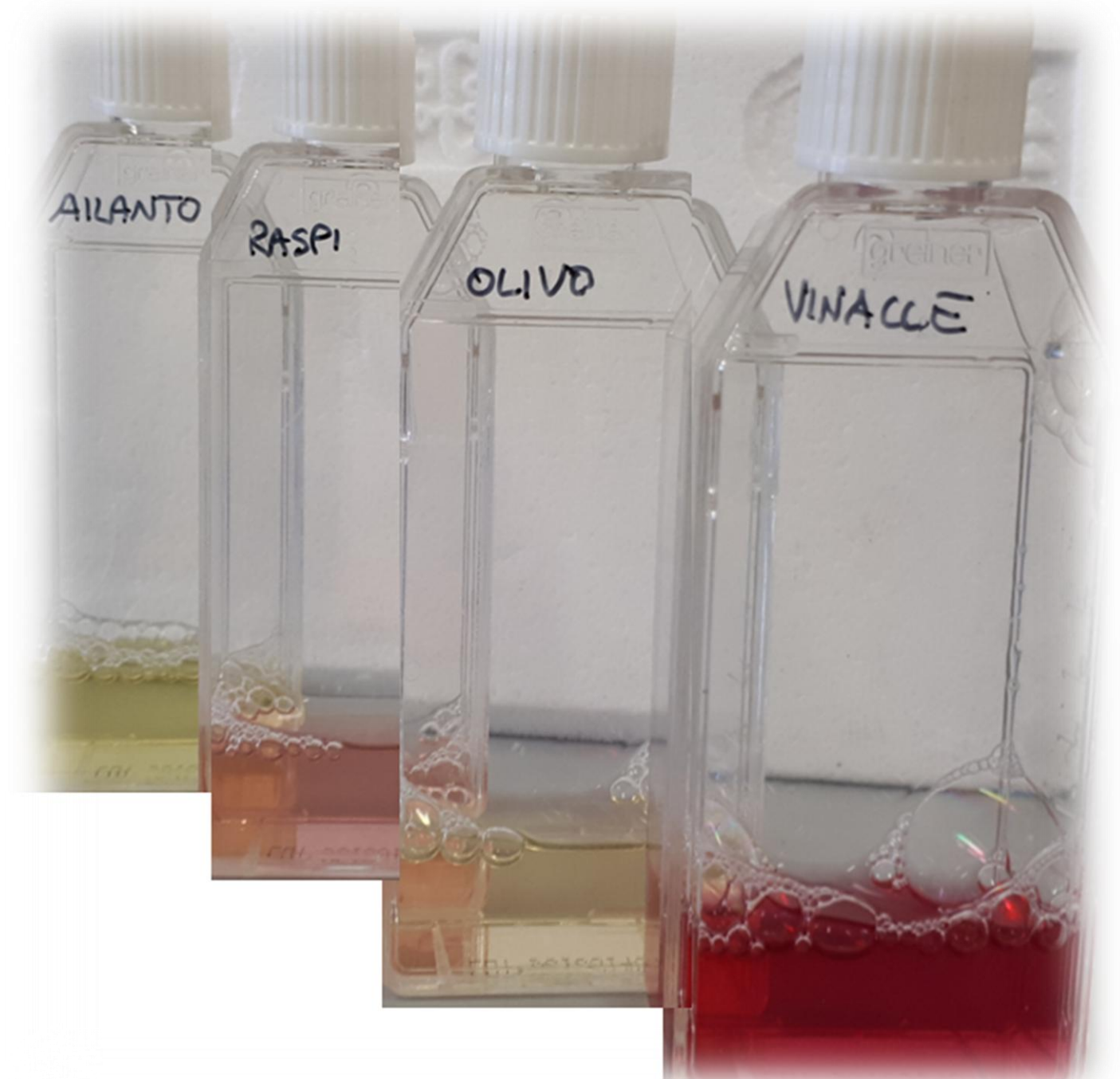
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### INTRODUCTION

Secondary metabolites confer a first set of defenses against pathogens and herbivore attack. The use of plant extracts (PE), enriched in these compounds, could represent an additional practice in environment friendly grapevine pest management. During biotic stress, the induction of different pathogenesis-related (PR) gene-expression, such as chitinase, often occurs. In the present work, four different plant extracts were tested for their modulation of chitinase activity in either grapevine suspension cell cultures (GSC) and plants.



### MATERIAL AND METHODS

**BA extracts:** Olive and Ailanthus leaves, grapevine marc and stalks (cv. Refosco dal peduncolo rosso) were extracted in 50% ethanol (v/v). PE were freshly diluted to concentration 4 and 800 g/L. **Experiments with grapevine cell suspension cultures:** GSC (cv. Cabernet Sauvignon) were grown as described in Krisa *et al.* [1999]. Seven independent replicates of 7-day old cells were incubated for 48 h with 4 g L<sup>-1</sup> of the four PE; chitinase activity were analysed as in Filippi *et al.* [2016]. Cell death was measured by fluorescein diacetate assay and counting at least 100 cells for each replicate. **Experiments with grapevine potted vines:** Two-year-old potted grape plants (cv. Verduzzo friulano) were grown in greenhouse at 25°C, 60% humidity, light/dark cycle of 14/10 h. At the stage of 7<sup>th</sup> fully expanded leaf, three plants per treatment were sprayed with 5 mL/plant of two different concentrations of grapevine marcs (4 and 800 g/L). After 8, 24 and 48 h after treatment, leaf chitinase activity was estimated.

### RESULTS AND DISCUSSION

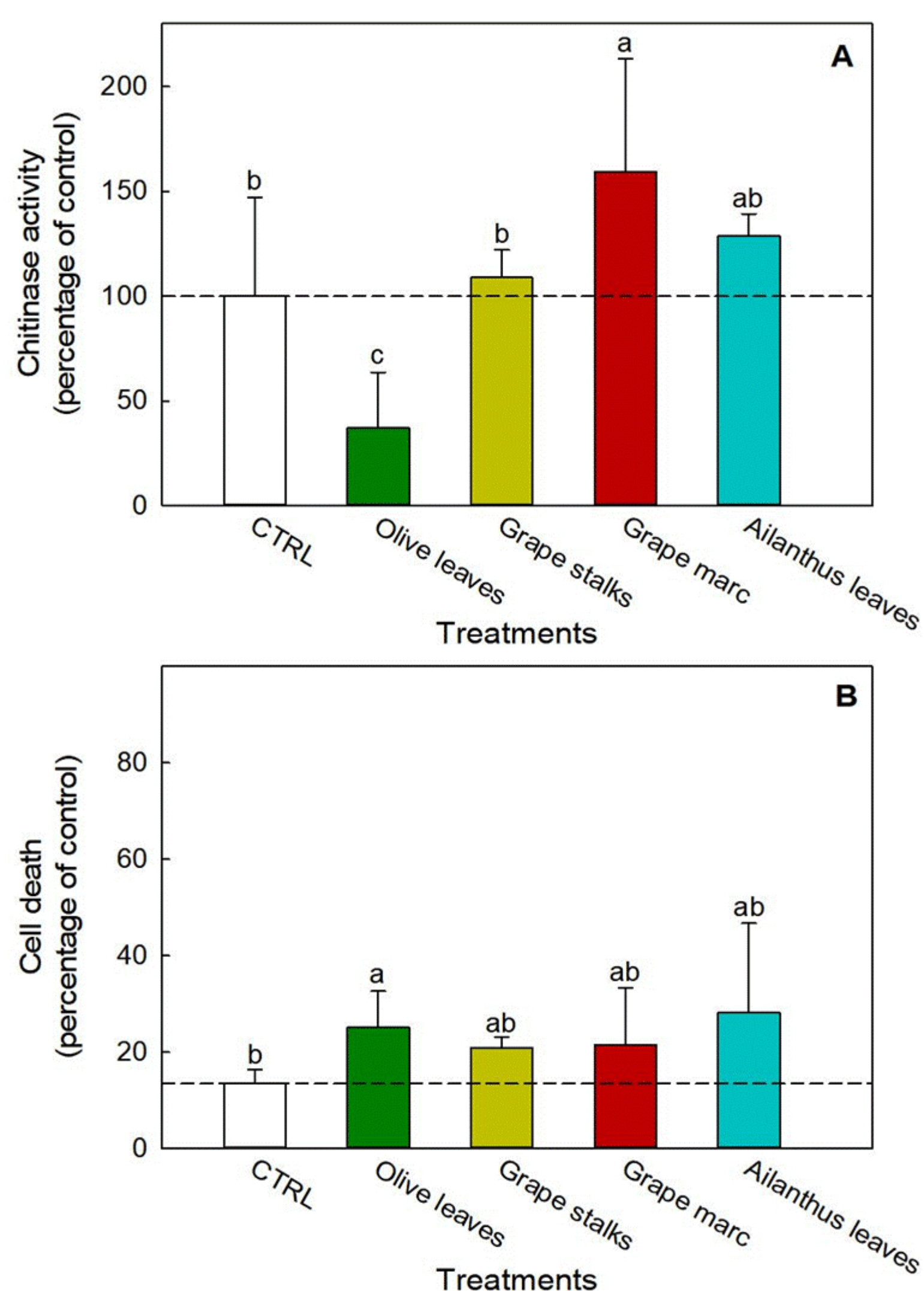


Fig. 1

**FIGURE 1**  
**Chitinase activity in GSC**  
Grapevine marc and olive leaves PE (4 g L<sup>-1</sup>) were able to increase chitinase activity by 1.5 fold or reduce to 50% of the control, respectively. No evident cytotoxic action were observed by evaluating the cell death (Panel B).

**FIGURE 2**  
**Chitinase activity in grapevine plants**  
The best PE (grapevine marc) was then chosen for further investigation in foliar application to potted vines. Control samples showed constitutive chitinase activity, which was nonetheless stimulated by grape marc at 4 g L<sup>-1</sup> over 24 or 48 h from treatment. The higher dose of 800 g L<sup>-1</sup> strongly stimulated foliar chitinase and the effect was anticipated even at 8 h after treatment.

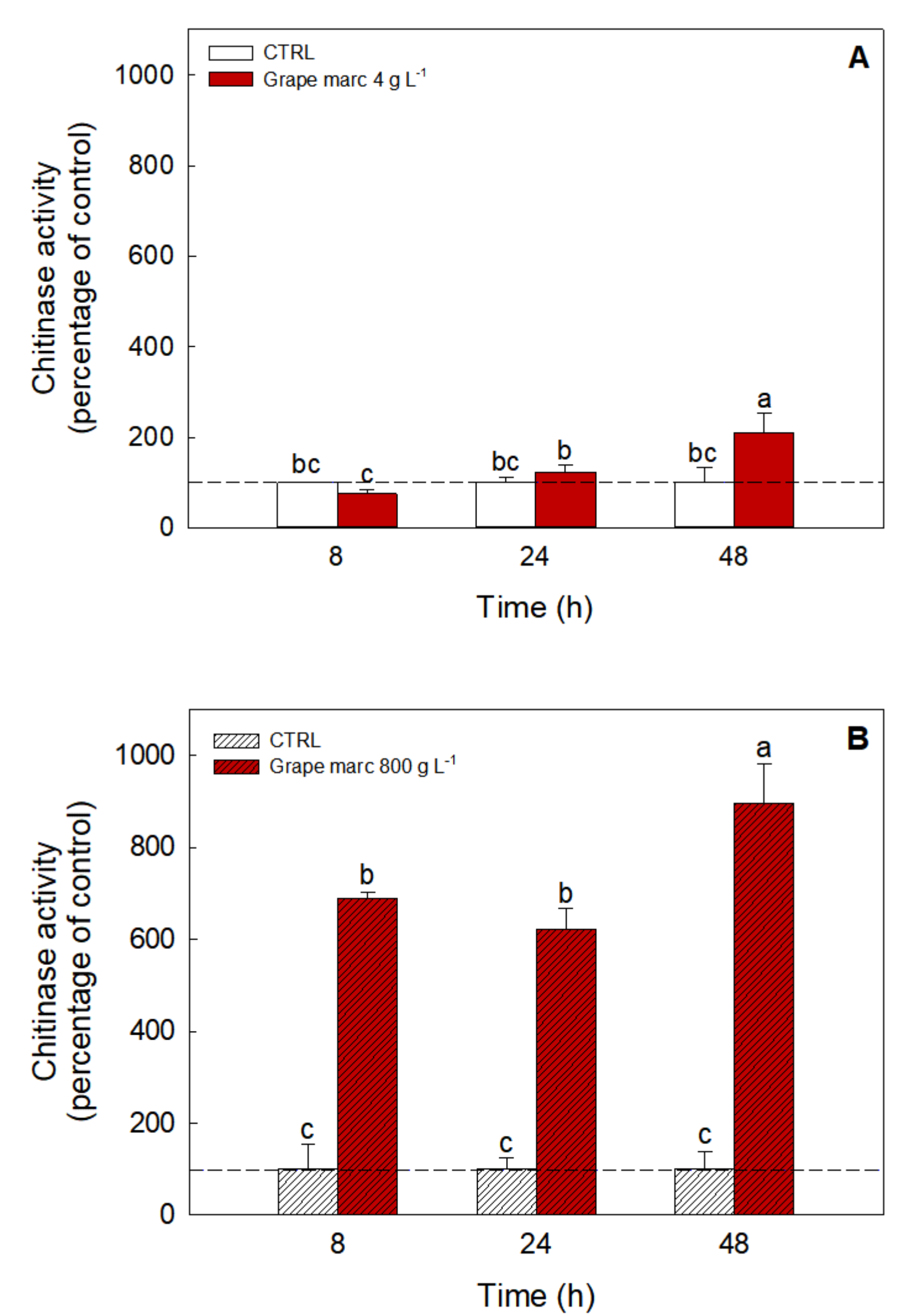


Fig. 2

### CONCLUSIONS

Our results demonstrate the **positive modulatory effect of grapevine marc PE**, applied on both cell cultures and grape plants. Grapevine chitinase represents one of the main players in the first line of defense against pathogen (Enoki & Suzuki, 2016). The assayed PE could thus be used as bioagent for **stimulating the vine immune resistance system**, in integration with other strategies undertaken for pest reduction in vineyard

### REFERENCES

Filippi *et al.*, (2016) Flavonoid Interaction with a chitinase from grape berry skin: protein identification and modulation of the enzymatic activity. *Molecules*. 21(10), 1300 doi.org/10.3390/molecules21101300  
Krisa *et al.*, (1999) Production of 13C-labelled anthocyanins by *Vitis vinifera* cell suspension cultures. *Phytochemistry*. 51:651-656 doi.org/10.1016/S0031-9422(99)00068-0  
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